

87. (Once Amended) The probe set of claim [20] 21, wherein the enzyme is selected from the group consisting of: a polymerase, alkaline phosphatase, horseradish peroxidase and soy bean peroxidase.

(iii) *Comments To The Amendment:*

Claims 9, 20, 33, 60 and 72 have been canceled as the limitations of said claims have been incorporated into claims 10, 21, 34, 61 and 80, respectively. Several other claims have been amended to correct for dependencies. Unmarked (clean) claims are set forth in Appendix A. Claim 80 has also been amended to more distinctly claim the subject matter for which Applicants seek letters patent. No new matter has been added by this amendment.

IV. RESPONSE TO REJECTIONS UNDER 35 U.S.C. 112, SECOND PARAGRAPH

The Examiner has reiterated, from the prior Office Action, the rejection of claims 46-49, 60-62 and 80-85 under 35 U.S.C. §112, second paragraph as being indefinite over recitation of the phrase "suitable *in-situ* hybridization conditions". In response to the arguments submitted on January 17, 2002, the Examiner has basically argued that the specification is lacking in providing the relevant information. In particular, the Examiner again argues that: "... **the specification does not provide a complete and fixed definition for this phrase**". Applicants continue to traverse this rejection.

It is well settled law that a claim is sufficiently definite for purposes of the second paragraph of 35 USC § 112 if one of ordinary skill in the art would understand what is claimed when considered in light of the specification and the prior art. *Penda Corporation v. United States*, 29 Fed.Cl. at 554. There is no requirement that the specification list every parameter of operation, so long as those of ordinary skill in the art realize that the parameter may be readily obtainable *Id.* Put differently, claims are not considered in a vacuum but rather are considered in view of the specification as well as the level of skill in the art (M.P.E.P § 2173.02). Although the claims form the meets and bounds of that which is patented, there is no legal requirement that the specification provide a complete and fixed definition for any term or phrase. The only legal requirement is that the ordinary practitioner be able recognize and understand the

claimed subject matter. In this regard, the Examiner should not object to claim language merely because he/she believes that more suitable language exists (M.P.E.P. § 2173.02).

It is respectfully submitted that the Examiner's analysis in maintaining this rejection fails to consider the claim language and the content of the specification when taken in view of the state of the art *in-situ* hybridization and the knowledge of the ordinary practitioner. It is respectfully submitted that the field of *in-situ* hybridization is well established and that the ordinary practitioner would understand the meets and bounds of the claims, particularly in the context of adjusting hybridization conditions so as to achieve an assay that performs acceptably for the purpose for which it operates.

In the present case, the Examiner objects to use of the phrase "suitable *in-situ* hybridization conditions". Claim 46, for example, clearly pertains to performing an *in-situ* assay since it is a requirement that the probe/target sequence hybrid form **within** the yeast and the enzyme activity is detected **within** the yeast. Perhaps the Examiner is arguing the assay might otherwise be successfully performed under **non-suitable** *in-situ* hybridization conditions. It is not likely that an ordinary practitioner would perform an *in-situ* assay under **non-suitable** *in-situ* hybridization conditions and expect to obtain a useful result. Accordingly, the specification describes the considerations that an ordinary practitioner might apply to determining "suitable *in-situ* hybridization conditions". Additionally, the Examples provide specific "suitable *in-situ* hybridization conditions" that were actually used in an exemplary assay. It is respectfully submitted that the ordinary practitioner would be able to exercise routine experimentation, given the information provided in the application (in addition to the knowledge expected for such an ordinary practitioner), to determine "suitable *in-situ* hybridization conditions" for each assay.

In support of the premise that the field of *in-situ* analysis is well established and that the ordinary practitioner would possess such knowledge and expertise, a list of recent papers published on using PNA and *in-situ* hybridization is attached hereto as Appendix B (**no publication being admitted as being prior art and in fact many of which are clearly not prior art**). It is respectfully submitted that the papers establish a level of skill attributable to the ordinary practitioner with regard to *in-situ* analysis that demonstrates that the claims at issue are neither vague nor indefinite under 35 U.S.C.

§112, second paragraph. Therefore, it is believed that the present rejection is improper and should be withdrawn.

Notwithstanding the foregoing, it is noted that claims 46-49 and 80-85 differ from those of claims 60-62 because whilst the former use the phrase "suitable *in-situ* hybridization conditions" the latter use the phrase "suitable hybridization conditions". It is respectfully submitted that the distinction is not relevant to the nature of the rejection as it is believed that either phrase is clear and definite in view of the teachings of the specification and the knowledge of the ordinary practitioner.

With respect to the rejection of claims 80-82, the Examiner has argued that it is unclear as to whether the reagents of a)-h) are considered to be the "reagents or compositions necessary to perform the assay or whether the reagents are present in addition to the "reagents or compositions necessary to perform the assay". It is believed that the amendment to claim 80 renders moot the present rejection.

In view of the foregoing remarks and amendments, it is respectfully submitted that all of the presently applied rejections under 35 U.S.C. §112, second paragraph should properly be withdrawn.

V. RESPONSE TO REJECTIONS UNDER 35 U.S.C. § 103(a)

(i) *The Facts*

- The Examiner acknowledges that Kosse does not teach enzyme-linked probes for the analysis of yeast, including *Dekkera bruxellensis* (OA at page 6).
- The Examiner acknowledges that Kosse teaches *in-situ* assays using fluorescently labeled probes whilst teaching dot-blot assays using digoxigenin labeled probes (OA at pages 5 and 6).
- Kosse teaches that only the 3' end of the 18S rRNA is accessible to fluorescently labeled probes and that the other variable regions of 18S rRNA were not accessible to *in-situ* hybridization. (Abstract and page 478, col. 2, first full paragraph).
- Kosse specifically describes the importance of permeabilization of the cell wall of yeast else probes will not penetrate and the yeast cannot be determined (Kosse at page 474, col. 1, first full paragraph). Kosse

specifically describes treatment of the yeast cells with **lyticase** to permeabilize the cells to the fluorescently labeled probes. *Id.*

- Stender (1998) does not teach anything about yeasts but is limited to determinations of mycobacteria.
- Stender (1998) teaches enzyme-linked probes for the determination of mycobacteria but does not teach about permeabilizing yeast cells to enzyme-linked probes.
- Stender (1998) did not actually use enzyme-linked probes in any assay and does not appear to have appreciated the difficulty that Amann et al. describe with regard to getting large probes (e.g. enzyme-linked probes) into cells having a cell wall.
- Amann et al. (Reference CA) specifically teach that enzyme-linked probes WOULD NOT penetrate into yeast cells (Abstract and pages 3008-3010, section entitled "Penetration of HRP-labeled oligonucleotides into whole fixed cells). Amann et al. explain that with a horseradish peroxidase label, the oligonucleotide probe is approximately 100 times larger than is a fluorescently labeled probe (Amann et al. at page 3008, bottom of col. 2). Amann et al. also teach that cells of *Saccharomyces cerevisiae* (a yeast) were not determinable with enzyme-linked probes even when treated with enzymes (**lyticase** and β -glucuronidase) or detergents (Amann at page 3010, middle, col. 1). It is noted that **lyticase** is the same enzyme that Kosse used to permeabilize yeast to fluorescently labeled oligonucleotide probes and Amann et al. specifically teach that said **lyticase** enzyme does not work to permeabilize yeast to enzyme-linked oligonucleotide probes. Stender (1998) is silent to this particular issue.

(ii) *Rejection based upon Kosse and Stender (1998)*

At paragraph 5 of the present Office Action, the Examiner has rejected claims 1-9, 12, 20, 23, 24, 25, 26, 29, 32, 33, 46, 60, 72, 86 and 87 under 35 U.S.C. §103(a) as being unpatentable over Kosse in view of Stender (1998). Applicants respectfully traverse this rejection.

Notwithstanding the rebuttal arguments the Examiner presents at pages 7-9 of the present Office Action, it is respectfully submitted that based upon the facts set forth above, the ordinary practitioner is without any proper motivation or reasonable expectation of successfully applying an enzyme-linked *in-situ* probe for the determination of yeast. Amann et al. is the most specific teaching on this subject matter and it clearly teaches away from such probe, probe set or method since they specifically teach that yeast cannot be determined *in-situ* using enzyme-linked probes. Neither of Stender (1998) nor Kosse provides any facts that undermine these specific teachings of Amann et al. Therefore Applicants stand ready to appeal this rejection as the combination of Kosse with Stender (1998) simply is not proper in view of the specific teachings of Amann et al. However, the following specific comments should also be considered with regard to the patentability of certain claims.

Regarding claim 1, and its dependent claims, it is respectfully submitted that the claim recites "An enzyme-linked *in-situ* hybridization probe..." and not "a probe suitable for *in-situ* hybridization" as the Examiner appears to suggest at page 8 of the Office Action.

Regarding claims 9, 20, 33, 60 and 72, these claims have been canceled and accordingly, it is believed that the rejection of these claims is rendered moot. Similarly, the dependencies of claims 12, 23, 34, 25, 26, 29, 32, 86 and 87 have been changed and therefore the rejection of these claims is believed to be rendered moot.

Regarding claim 46, and claims dependent thereon, as argued above, clearly this claim pertains to an *in-situ* assay since the one or more probe/target sequences are formed **within** the yeast and the enzyme activity is detected **within** the yeast. For reasons stated above, Amann et al. clearly teach away from such an assay. Thus, it is believed that claim 46 is both novel and non-obvious over Kosse in view of Stender (1998).

Regarding claims 10, 21, 34 and 61, it is believed that Kosse and Stender (1998), alone or in combination, do not teach the sequences described in the claims. Accordingly, it is believed that these claims are both novel and non-obvious over the cited references.

In view of the foregoing remarks, as well as the amendment set forth herein, it is respectfully submitted that the present rejection of claims under 35 U.S.C. §103(a) over Kosse in view of Stender (1998) should be withdrawn. Reconsideration is requested.

(ii) *Rejection based upon Kosse in view of Stender (1998) and Parton (5,905,038)*

At paragraph 6 of the present Office Action, the Examiner has rejected claims 47-49 and 80-85 under 35 U.S.C. §103(a) as being unpatentable over Kosse in view of Stender (1998) and further in view of Parton (US 5,905,038). Applicants respectfully traverse this rejection.

For reasons described above, it is believed that claim 46 is both novel and non-obvious. Accordingly, dependent claims 47-49 are believed to be patentable as being dependent upon an allowable base claim.

Regarding claims 80-82, it is believed that the amendment to claim 80 renders moot the rejection of claims 80-82.

Regarding claims 83-85, it is respectfully submitted that the references, alone or in combination, do not describe the determination of microcolonies of slow growing yeast. Accordingly, the combined references do not teach all of the elements/limitations of claims 83-85.

In view of the foregoing remarks, as well as the amendment set forth herein, it is respectfully submitted that the present rejection of claims under 35 U.S.C. §103(a) over Kosse in view of Stender (1998) and in further view of Parton should be withdrawn. Reconsideration is requested.

(iii) *Rejection based upon De Wachter in view of Kosse and Stender (1998)*

At paragraph 7 of the present Office Action, the Examiner has rejected claims 1-12, 16, 18-26, 29, 32, 33, 46, 60-62, 72, 86 and 87 under 35 U.S.C. §103(a) as being unpatentable over De Wachter in view of Kosse and in further view of Stender (1998). Applicants respectfully traverse this rejection.

It is respectfully reiterated that Kosse and Stender (1998) are not properly combined for reasons previously stated. Moreover, De Wachter is merely a disclosure of a gene sequence and not the rRNA sequence itself. Kosse, however, specifically teaches that only the 3' end of the 18S rRNA is accessible to fluorescently labeled probes

and that the other variable regions of 18S rRNA were not accessible to *in-situ* hybridization. Consequently, it is not reasonable to expect any nucleobase sequence that is homologous to the gene sequence described by De Wachter will be useful to produce an *in-situ* hybridization probe. This express teaching of Kosse expressly undermines the Examiner's statement that: "... DeWachter is considered to have the property of being suitable as a probe for the detection, identification or quantitation of *Dekkera/Brettanomyces bruxellensis*." As the premise of the Examiner's rejection is undermined, the rejection should not properly stand.

In view of the foregoing remarks, as well as the amendment set forth herein, it is respectfully submitted that the present rejection of claims under 35 U.S.C. §103(a) over De Wachter in view of Kosse and Stender (1998) should be withdrawn. Reconsideration is requested.

(iv) Rejection based upon De Wachter in view of Kosse and Stender (1998) and further in view of Parton

At paragraph 8 of the present Office Action, the Examiner has rejected claims 47-49 and 80-85 under 35 U.S.C. §103(a) as being unpatentable over De Wachter in view of Kosse and Stender (1998) in further view of Parton (US 5,905,038). Applicants respectfully traverse this rejection.

For reasons discussed above, it is believed that Kosse and Stender are not properly combined. Moreover, it is believed that De Wachter and Kosse are not properly combined for reasons previously stated. Finally, Parton does not cure any of these deficiencies.

In view of the foregoing remarks, as well as the amendment set forth herein, it is respectfully submitted that the present rejection of claims under 35 U.S.C. §103(a) over De Wachter in view of Kosse, Stender (1998) and Parton should be withdrawn. Reconsideration is requested.

VI. SUMMARY

It is believed that this response addresses all rejections set forth in the present Office Action and the application is in ready condition for allowance. In consideration of the preceding amendments and remarks, Applicants hereby respectfully request

reconsideration of all pending claims (as amended herein), the withdrawal of all rejections set forth in the present Office Action and issue of a Notice of Allowance by The Office.

VII. INTERVIEW

If the Examiner believes a telephonic or personal interview would advance the prosecution of the subject application, the Examiner is invited to contact attorney Gildea during business hours at the telephone or facsimile numbers listed below.

VIII. FEES

Except for the fee due for consideration of the petition under 37 C.F.R. §1.136(a), it is believed that no additional fees are believed due The Office for consideration of this paper. If however, The Office determines that any other fee is due, authorization is hereby granted to charge any required fee associated with the filing and consideration of this paper to Deposit Account 02-3240.

IX. CORRESPONDENCE/CUSTOMER NUMBER

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Respectfully submitted
on behalf of Applicants,

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